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NEGATIVE SELECTION EFFECTS SUPPRESS RELATIONSHIPS BETWEEN BACTERIAL DIVERSITY AND ECOSYSTEM FUNCTIONING

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Abstract. I assembled bacterial communities to explore the effects of bacterial diversity on multiple ecosystem functions, including bacterial community biovolume, decomposition of particulate organic matter, and biomass transfer to the next trophic level. The experiment used a two-way factorial design with four levels of bacterial diversity (one to four species) and the absence/presence of a bacterivorous ciliated protist *Tetrahymena pyriformis* as two main factors, and all possible combinations of the four bacterial taxa nested within each diversity level. Bacterial community biovolume increased as bacterial diversity increased, a result due largely to positive selection effects. Decomposition and consumer abundance, however, were unaffected by bacterial diversity, though both varied among bacterial composition treatments. Negative selection effects, the dominance of species that do not contribute significantly to ecosystem functioning, accounted for the lack of diversity effects on decomposition and consumer abundance. The presence of *Tetrahymena* reduced bacterial community biovolume but increased decomposition, without altering the diversity–functioning relationships. Decomposition was strongly linked with consumer abundance such that communities supporting larger consumer biomass exhibited higher decomposition rates. This study suggests that if the negative selection effect is common, as it might be when examining ecosystem variables other than biomass (due to the presence of keystone species that can contribute disproportionably to ecosystem functioning relative to their abundances), basic bacteria-mediated ecosystem processes, such as decomposition and energy transfer to the next trophic level, may not increase with bacterial diversity.

Key words: aggregate community biomass; bacterial diversity; biomass transfer among trophic levels; decomposition; trophic interactions.

INTRODUCTION

The relationship between biodiversity and ecosystem functioning (hereafter BEF) has been the focus of much recent research, largely driven by the need to understand how ongoing and future changes in biodiversity may affect ecosystem processes, goods, and services. Accumulating theoretical and empirical evidence suggests a common positive BEF relationship (Kinzig et al. 2002, Loreau et al. 2002, Hooper et al. 2005, Srivastava and Vellend 2005, Cardinale et al. 2006). Positive effects of diversity on ecosystem processes have been attributed to two primary mechanisms: complementarity and positive selection (or sampling) effects. Complementarity develops when niche differentiation among species, which reduces interspecific competition, allows diverse communities to utilize available resources more completely (Tilman et al. 1997); positive selection effects develop when competition leads to dominance of species with strong influences on ecosystem processes and diverse communities have greater probabilities of containing these species (Aarssen 1997, Huston 1997, Tilman et al. 1997).

Although common, the positive BEF relationship is not universal. Notable exceptions include a number of experimental studies that showed little or no effect of plant diversity on primary production (Hooper and Vitousek 1997, Troumbis et al. 2000, Engelhardt and Ritchie 2002, Hooper and Dukes 2004). In these studies, a negative selection effect operated where superior competitors were not the most productive species when grown alone and thus species with lower primary production dominated in diverse communities; negative selection effects counteracted and sometimes over-

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whelmed any positive influence of diversity on primary production due to complementarity, resulting in the lack of diversity effect. Negative selection effects have also been documented in studies that reported positive plant BEF relationships (Loreau and Hector 2001, Spaekova and Leps 2001, Hector et al. 2002, Bruno et al. 2005, Spehn et al. 2005), where they were of smaller magnitude compared with complementarity effects. Constraints on resource allocation to different plant parts are thought to give rise to negative selection effects associated with primary production (Hector et al. 2002, Hooper and Dukes 2004). Although rarely explored, negative selection effects are probably more common when ecosystem properties other than biomass production are examined, because superior competitors, though often achieving large abundances, may not dominate other ecosystem functions and subordinate competitors, while not numerically dominant, may contribute significantly to other ecosystem processes. For example, Lyons and Schwartz (2001) showed that uncommon plant species offer important resistance to the invasion of exotic plants. Indeed, ecological communities often contain species with disproportionably large impacts on community and ecosystem properties relative to their abundances (keystone species, sensu Power et al. 1996).

As an integral component of virtually all ecosystems, bacteria perform important ecosystem functions: as decomposers they break down organic materials and recycle nutrients, and at the basal level of detrital food webs, they constitute principal food resources for many species, particularly heterotrophic flagellates and ciliates. Despite these important roles of bacteria, we know remarkably little about the functional significance of bacterial diversity. Several researchers have used fumigation or dilution methods to explore the functional consequences of losses of bacterial diversity (e.g., Griffiths et al. 2000, 2001), but diversity effects cannot be separated from species composition effects in these studies, making it difficult to generalize their results beyond the particular systems under investigation (Huston 1997). Only a handful of studies have to date directly manipulated bacterial diversity and composition that allowed more unambiguous evaluations of the effects of bacterial diversity on ecosystem functioning (Naeem et al. 2000, Hodgson et al. 2002, Wohl et al. 2004, Bell et al. 2005). Of these, only one study (Wohl et al. 2004) examined bacterial diversity effects on organic matter decomposition and one study (Naeem et al. 2000) examined bacterial diversity effects on carbon substrate utilization, and none considered the effects of bacterial diversity on energy transfer to higher trophic levels.

Here, I report the results of an experimental study investigating how changes in bacterial diversity affect the magnitude of multiple ecosystem functions, including decomposition of particulate organic matter, biomass transfer to the higher trophic level, and bacterial community biomass. While bacterial community biomass is generally not viewed as a relevant bacterial function, it is considered here to compare with studies conducted in other systems in which biomass production is often the focal ecosystem variable (see also Hodgson et al. 2002). This study aimed to answer three main questions. First, how do changes in bacterial diversity affect the functioning of bacterial communities? Second, what mechanisms, including complementarity, positive selection, and negative selection effects, can account for the observed BEF relationships? Third, does the presence of bacterivorous consumers affect the BEF relationship? Several experiments have shown that the addition of higher trophic level organisms altered the form of BEF relationships in plant or algal-based systems (e.g., Mulder et al. 1999, Duffy et al. 2005), but whether these findings can be extended to detritus-based microbial communities remains unclear.

Materials and Methods

Because many previous experiments found the strongest effects of diversity on ecosystem functioning at low diversity levels (e.g., Tilman et al. 2001, Bell et al. 2005) and because adequate species composition treatments are necessary for assessing mechanisms underlying biodiversity effects (Allison 1999, Schmid et al. 2002), I used a small bacterial diversity gradient (one to four species) with every possible composition within each diversity level represented and replicated. The four bacterial species were isolated from the Rutgers University Display Garden pond (New Brunswick, New Jersey, USA; see Plate 1) and identified as *Bacillus cereus, Bacillus pumilus*, *Frigoribacterium* sp., and *Serratia marcescens* using sequence analysis of the 16s rRNA gene. Colonies of these four bacterial species can be readily distinguished on nutrient agar plates. A ciliated protist species, *Tetrahymena pyriformis*, was used as the consumer of bacteria. Axenic *Tetrahymena* cultures (grown in proteose peptone medium) were obtained from Carolina Biology Supply Company (Burlington, North Carolina, USA). Prior to the experiment, bacterial stock cultures were established by growing each species in 0.8% nutrient broth.

The experiment used a two-way factorial design with bacterial diversity and the presence/absence of *Tetrahymena* as the two main factors. Bacterial diversity consisted of four levels: one, two, three, and four species, with every possible species combination included within each diversity level. Each species composition was replicated three times, for a total of 90 experimental units (microcosms).

Each microcosm consisted of a 25-mL plug-seal-capped test tube containing 10 mL of nutrient medium. The nutrient medium was made by mixing 0.367 g protozoan pellet (Carolina Biological Supply Company, Burlington, North Carolina, USA) and 1.67 g garden soil in 1 L of deionized water. This medium was autoclaved and filtered to remove undissolved particles, before being transferred into experimental microcosms. Microcosms containing medium were again autoclaved.

At the start of the experiment (day 0), I inoculated microcosms with desired bacterial species by transferring a small volume (<0.5 μL) of bacterial stock cultures with an inoculating loop. As bacterial species differed in their stock culture density (indicated by differences in medium turbidity), and the transferred volume was the same for each species in all treatments, initial total bacterial density varied among bacterial diversity and composition treatments. Considering the short generation times of bacteria, however, this variation should not affect experimental results obtained after numerous bacterial generations (total experimental duration was 44 days). I introduced a small number of the bacterivorous consumer *Tetrahymena* into their designated microcosms on day 7, when bacterial densities had reached a stationary phase. To measure decomposition of particulate organic matter, I added one wheat seed into each microcosm seven days after *Tetrahymena* introduction (day 14). Prior to the experiment, wheat seeds were boiled, dried, weighed and autoclaved. Microcosms were placed on a shaker at 200 rpm and incubated at room temperature during the experiment.

I sampled microcosms at the end of the experiment (day 42 and 43) to quantify bacterial density, consumer abundance, and decomposition. Densities of each bacterial species were determined by plating serially diluted samples onto nutrient agar plates and counting the number of colonies at appropriate dilution levels after six days' incubation. Microscopic examination indicated that cells of the four bacterial species, which were all rod shaped, differed in size. For each bacterial species, I obtained its average individual biovolume by measuring the length and width of ten haphazardly selected acridine orange-stained cells under epifluorescence at 1000×. Bacterial community biovolumes were obtained by summing population biovolumes (average individual biovolume × population density) across all species in the community. *Tetrahymena* population densities were determined by counting the number of individuals present in 50-μL samples at 25×; samples containing dense populations were diluted before counting. Large variation in *Tetrahymena* body size among bacterial composition treatments was noted during counting, making it necessary to measure *Tetrahymena* body size. I then haphazardly chose three *Tetrahymena* individuals from each replicate (nine individuals for each bacterial composition treatment) and measured their cell length and width at 400×. *Tetrahymena* individual biovolume was calculated with a formula that approximates the cell shape (Wetzel and Likens 2000). Consumer biomass was indexed by *Tetrahymena* population biovolume (average individual biovolume × population density). Wheat seeds retrieved from microcosms were oven dried and weighed. Decomposition was defined as the fraction of wheat seed mass loss.

Unless otherwise stated, biovolume data were $\log_{10}$-transformed ($\log_{10}[x + 1]$) before analyses to improve normality. Analysis of covariance (ANCOVA), with bacterial diversity as the continuous variable and the presence/absence of *Tetrahymena* as the class variable, was used to test for the general trend of responses of bacterial community biovolume and decomposition to changes in bacterial diversity. Two-way ANOVAs, with bacterial composition and the presence/absence of *Tetrahymena* as the two main factors, were also used to test for the effect of bacterial composition and predation on the same ecosystem variables. Linear regression and one-way ANOVA were used to examine the effects of bacterial diversity and composition on *Tetrahymena* abundance, respectively. To assess possible biodiversity mechanisms (i.e., complementarity and selection effects) at work, the overyielding index $D_{\max}$, which compares the magnitude of ecosystem functioning of a polyculture with that of the constituent species with best-performing monocultures (Loreau 1998), was calculated for each polyculture. The additive partition model of Loreau and Hector (2001) cannot be used here because initial species abundances, key variables of the model, generally have minimal effects on outcomes of such long-term (in terms of generation times) experiments. The impossibility of measuring contributions of individual bacterial species to decomposition and supporting consumer populations in bacterial polycultures also prevented the use of the additive partition model or other commonly used indices (e.g., relative yield total [Loreau 1998]). While positive $D_{\max}$ indicates a complementarity effect and $D_{\max}$ close to zero indicates a positive selection effect (Loreau 1998), significantly negative $D_{\max}$, together with the observation that species important for the focal ecosystem variable reached substantially lower abundances in polycultures than monocultures, suggests a negative selection effect. Note that as a relatively conservative test for complementarity, $D_{\max}$ may not be able to detect complementarity effects of substantially smaller magnitude than selection effects; it, however, provides a reasonably quantitative measurement on their relative importance. I obtained 95% bootstrap confidence intervals of $D_{\max}$ by resampling the monoculture and polycluture data with replacement 500 times, and calculating $D_{\max}$ by pairing the resampled monoculture and polyculture replicates. All $D_{\max}$ values were calculated using untransformed data.

Results

Bacterial community biovolume increased as bacterial diversity increased (Fig. 1A; ANCOVA, diversity, $F_{1,86} = 16.75$, $P < 0.0001$), and the presence/absence of *Tetrahymena* did not affect this increasing trend (ANCOVA, diversity × predation, $F_{1,86} = 1.34$, $P = 0.2511$). *Tetrahymena* predation significantly reduced bacterial community biovolume, by approximately one order of magnitude at each diversity level (Fig. 1A; predation: $F_{1,86} = 4.01$, $P = 0.0484$). Bacterial community biovolume differed markedly among bacterial

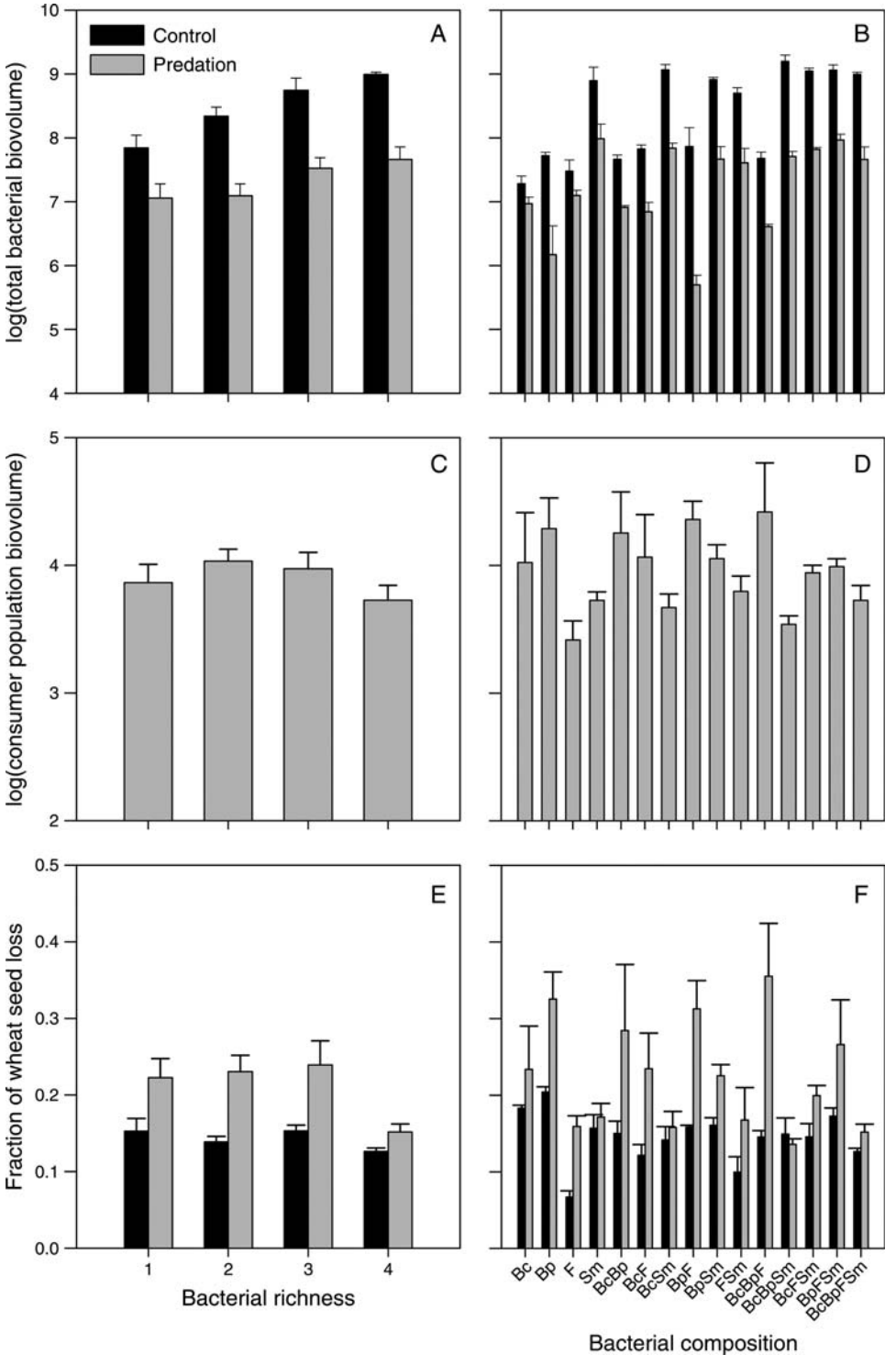

FIG. 1. Effects of bacterial diversity (left column) and composition (right column) on (A and B) bacterial community biovolume, (C and D) consumer population biovolume, and (E and F) decomposition of wheat seeds, under control and predation treatments. Error bars represent +SE. Abbreviations: Bc, *Bacillus cereus*; Bp, *Bacillus pumilus*; F, *Frigoribacterium* sp.; Sm, *Serratia marcescens*. Log-transformed bacterial and consumer biovolumes were originally measured as $\mu m^3/mL$. Bacterial richness refers to the number of species.

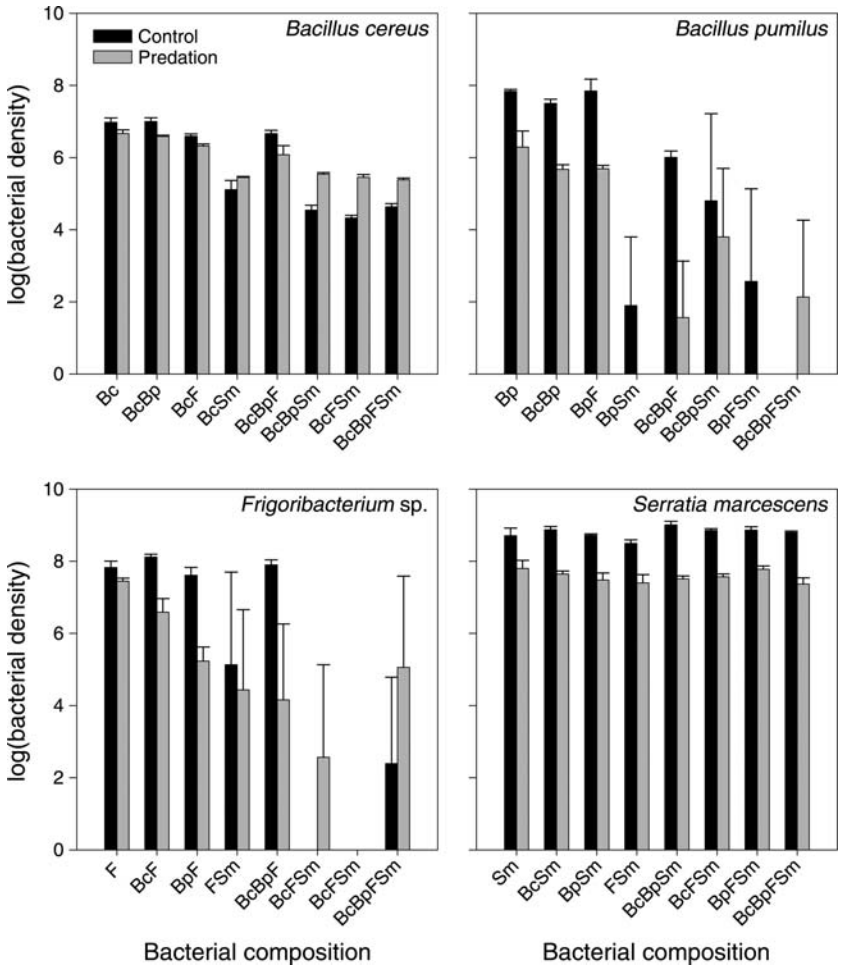

FIG. 2. Individual bacterial densities with different species composition treatments under control and predation treatments. Abbreviations: Bc, *Bacillus cereus*; Bp, *Bacillus pumilus*; F, *Frigoribacterium* sp.; Sm, *Serratia marcescens*. Log-transformed bacterial density was originally measured as CFUs/mL (CFUs, colony-forming units). Error bars represent + SE.

composition treatments (Fig. 1B, two-way ANOVA, composition: $F_{14,60} = 37.78$, $P < 0.0001$). In both control and predation treatments, *S. marcescens* was most productive in monocultures and dominated polycultures whenever present (Fig. 2). In the predator-free controls, except for two treatments (*B. cereus–Frigoribacterium* and *B. cereus–Bacillus pumilus–S. marcescens*) that exhibited significant positive D_{max} values suggestive of complementary interactions, other D_{max} values did not deviate from zero (Table 1). In the predation treatments, while most D_{max} values again did not deviate from zero, two treatments (*B. pumilus–Frigoribacterium* and *B. cereus–B. pumilus–Frigoribacterium*) showed significantly negative D_{max} values (Table 1). Negative D_{max} resulted from negative selection effects where *Frigoribacterium* sp., which was most productive in monocultures among *B. cereus*, *B. pumilus*, and *Frigoribacterium* sp. in the presence of *Tetrahymena*, attained substantially lower abundances in polycultures (Fig. 2). Apparent competition was largely responsible for this pattern. For instance, in the *B. pumilus–Frigoribacterium* treatment, *B. pumilus*, which was little affected by *Frigoribacterium* sp., supported higher *Tetrahymena* biomass than can be supported by *Frigoribacterium* sp. alone (Fig. 1D), which in turn reduced the abundance of *Frigoribacterium* sp. (Fig. 2). A similar pattern occurred in the *B. cereus–B. pumilus–Frigoribacterium* treatment.

Changes in bacterial diversity did not affect *Tetrahymena* abundance (Fig. 1C; linear regression, $R^2 = 0.0000$, $P = 0.9908$). Different bacterial composition treatments, however, showed marginally significant differences in *Tetrahymena* abundance (Fig. 1D, ANOVA, $F_{14,30} = 2.01$, $P = 0.0533$). Species compositions that supported the largest *Tetrahymena* populations include monocultures of *B. pumilus* and polycultures containing *B. pumilus* but not *S. marcescens* (Fig. 1D). *Tetrahymena* was less abundant in polycultures containing both *S. marcescens* and *B. pumilus* than monocultures of

TABLE 1. Bootstrapped 95% confidence intervals (CI) of D_{max} (overyielding index) associated with bacterial community biovolume, consumer population biovolume, and wheat seed decomposition.

Species composition	Bacterial community biovolume CI		Consumer population biovolume CI	Decomposition CI	
	Predation-free controls	Predation treatments		Predation-free controls	Predation treatments
BcBp	−0.332 to 0.195	−0.355 to 0.201	−0.718 to 2.527	**−0.418 to −0.159**	−0.590 to 0.334
BcF	**0.380 to 3.033**	−0.677 to 0.222	−0.683 to 16.373	**−0.439 to −0.181**	−0.351 to 1.018
BcSm	−0.294 to 2.490	−0.669 to 1.084	−0.868 to 1.862	**−0.384 to −0.054**	−0.532 to 0.158
BpF	−0.435 to 4.233	**−0.977 to −0.923**	−0.289 to 2.167	**−0.264 to −0.179**	−0.246 to 0.335
BpSm	−0.426 to 1.119	−0.771 to 0.615	−0.692 to 0.456	**−0.299 to −0.124**	**−0.428 to −0.128**
FSm	−0.700 to 0.610	−0.793 to 0.939	−0.218 to 0.864	**−0.539 to −0.114**	−0.347 to 0.647
BcBpF	−0.362 to 0.423	**−0.761 to −0.577**	−0.704 to 4.578	**−0.368 to −0.218**	−0.273 to 0.579
BcBpSm	**0.017 to 4.565**	−0.748 to 0.520	**−0.885 to −0.623**	**−0.399 to −0.037**	**−0.641 to −0.477**
BcFSm	−0.238 to 2.228	−0.629 to 0.592	−0.735 to 3.253	**−0.388 to −0.083**	−0.373 to 0.458
BpFSm	−0.244 to 2.681	−0.567 to 1.848	**−0.676 to −0.005**	**−0.263 to −0.058**	−0.451 to 0.189
BcBpFSm	−0.319 to 1.570	−0.780 to 0.568	**−0.855 to −0.392**	**−0.426 to −0.335**	**−0.607 to −0.429**

Notes: D_{max} values that differ significantly from 0 are shown in boldface type. Abbreviations: Bc, *Bacillus cereus*; Bp, *Bacillus pumilus*; F, *Frigoribacterium* sp.; Sm, *Serratia marcescens*.

B. pumilus (with the exception of the *B. pumilus–S. marcescens* treatment), as *B. pumilus* declined drastically in abundance in the presence of *S. marcescen* (Fig. 2), resulting in negative selection effects indicated by negative D_{max} values in these treatments (Table 1). D_{max} did not differ from zero in other treatments.

Decomposition was also unaffected by changes in bacterial diversity, regardless of the presence/absence of *Tetrahymena* (Fig. 1E; ANCOVA, diversity, $F_{1,86} = 0.35$, $P = 0.5554$; diversity × predation, $F_{1,86} = 0.05$, $P = 0.8240$). *Tetrahymena* predation had a significantly positive effect on decomposition, which, with the exception of the four-species treatment, showed approximately 50% increase in the presence of *Tetrahymena* (Fig. 1E; ANCOVA, predation: $F_{1,86} = 5.13$, $P = 0.0260$). There was large variation in decomposition among monocultures, with *B. pumilus* being the best decomposer in both control and predation treatments. Polycultures containing *B. pumilus* but not dominant *S. marcescens* also showed elevated decomposition values above other treatments in the presence of consumers (Fig. 1F). As a result, two-way ANOVA revealed a significant composition effect ($F_{14,60} = 4.32$, $P < 0.0001$), a significant predation effect ($F_{14,60} = 4.32$, $P < 0.0001$), and a marginally significant composition × predation effect ($F_{14,60} = 1.84$, $P = 0.0537$). In the predator-free controls, all D_{max} values were significantly less than zero, whereas in the predation treatments, negative D_{max} values were found for *B. pumilus–S. marcescens*, *B. cereus–B. pumilus–S. marcescens*, and *B. cereus–B. pumilus–Frigoribacterium–S. marcescens* treatments, with others treatments exhibiting values not significantly from zero (Table 1). The negative D_{max} values were primarily a result of the negative selection effect: good decomposers, such as *B. pumilus*, did not dominate numerically and often declined in abundance in polycultures (Fig. 2). Interestingly, decomposition was significantly and positively correlated with *Tetrahymena* abundance within the predation treatments: variation in *Tetrahymena* population biovolume explained 69% of the variation in decomposition (Fig. 3A). A similar pattern existed when only bacterial monocultures were considered (Fig. 3B).

DISCUSSION

This study shows that the form of the bacterial BEF relationship depends on the ecosystem variable examined. Bacterial community biovolume increased as bacterial diversity increased, whereas decomposition and consumer population biovolume showed little response to changes in bacterial diversity. Regardless of the form of the BEF relationship, the overyielding index, D_{max}, rarely attained positive values (D_{max} being significantly positive in only two of 55 cases, Table 1), suggesting that complementarity, if present, was generally insufficient to cause better polyculture performances over the corresponding monocultures. The small role of the complementarity effect runs counter to the conclusion of a recent study that complementarity was primarily responsible for the positive bacterial diversity effect on ecosystem respiration (Bell et al. 2005). Bell et al. (2005), however, reached their conclusion without directly testing for selection effects, and inspection of their experimental data reveals that maximum ecosystem respiration did not differ among diversity levels containing more than one species, a sign of the positive selection effect. The positive selection effect, that increasing diversity increased the probability of including the competitively dominant and productive species (*S. marcescens*), was thus largely responsible for the positive relationship between bacterial diversity and community biovolume. This result agrees with the finding of a recent meta-analysis of 111 BEF experiments that much of the positive effect of biodiversity on aggregate community biomass can be attributed to the positive selection effect (Cardinale et al. 2006).

Species with strong influences on ecosystem processes, however, may not always dominate mixtures. For

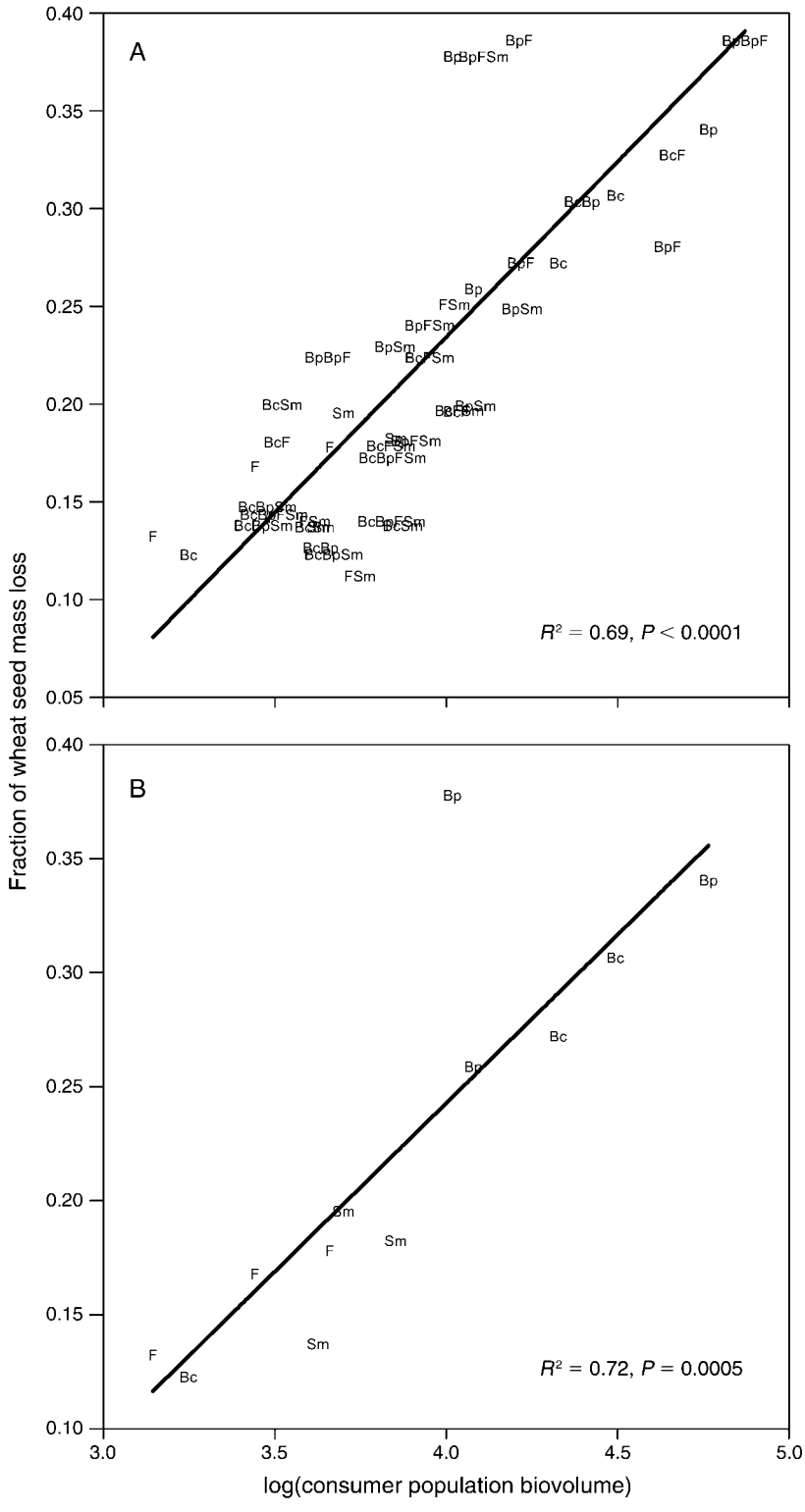

FIG. 3. The positive relationship between decomposition (fraction of wheat seed mass loss) and *Tetrahymena* population biovolume in the predation treatments for (A) data including monocultures and polycultures and (B) data including only monocultures. Abbreviations: Bc, *Bacillus cereus*; Bp, *Bacillus pumilus*; F, *Frigoribacterium* sp.; Sm, *Serratia marcescens*.

PLATE 1. The small pond located in the Rutgers University Display Garden, New Brunswick, NJ (USA), the collecting site of the four bacterial species. Photo credit: P. J. Morin.

example, plants allocating much of their acquired resources to aboveground biomass production may have shallow roots or low stature, rendering them weak competitors for nutrients or light. This constraint on resource allocation could generate a negative selection effect, as found by a number of plant BEF studies (Hooper and Vitousek 1997, Troumbis et al. 2000, Loreau and Hector 2001, Spaekova and Leps 2001, Engelhardt and Ritchie 2002, Hector et al. 2002, Hooper and Dukes 2004, Bruno et al. 2005, Spehn et al. 2005). As noted in the introduction, negative selection effects are probably more common for ecosystem functions other than aggregate community biomass, because it is not uncommon for ecological communities to contain species with disproportionately large influences on ecosystem processes relative to their abundances (Power et al. 1996). Consistent with this hypothesis, *B. pumilus*, a competitively subordinate species, supported the largest consumer abundance and was associated with the highest decomposition rate in monocultures. As a result, the negative selection effect emerged and dominated, resulting in consumer abundance and decomposition largely decoupled from bacterial diversity.

Only a few studies have directly examined the effect of biotic resource diversity on consumer abundance (Petchey 2000, Haddad et al. 2001, Fox 2004, Gamfeldt et al. 2005). Among these, Petchey (2000) and Gamfeldt et al. (2005) reported positive effects of resource diversity, and Haddad et al. (2001) and Fox (2004) reported negative and/or no effects of resource diversity. Differences in resource edibility appear to underlie these different patterns. The former studies used resource species that were of high edibility for consumers, and consumer abundance increased with resource diversity as resource abundance increased due to complementarity and/or positive selection effects or consumers benefited nutritionally from mixed resources (Petchey 2000, Gamfeldt et al. 2005). The latter studies included inedible or unpalatable resource species and competition from these species reduced the abundance of edible species in mixtures, resulting in negative selection effects. Consistent with the latter scenario, my experiment with bacterial species differing in palatability (Fig. 1D) found that changing bacterial diversity did not affect bacterivorous consumer abundance. Taken together, these results suggest that there may not be a general relationship between resource diversity and consumer abundance. One could, however, argue that a positive relationship between resource diversity and consumer abundance may not be common. The two experiments documenting a positive resource diversity effect used a limited number (up to three) of edible resource species (Petchey 2000, Gamfeldt et al. 2005); given that heterogeneity in species edibility generally characterizes ecological communities (e.g., Paine 1966, Lubchenco 1978, McNaughton 1983, Leibold 1989, Jiang and Morin 2005), further increases in diversity will increase the chance of including less-edible species.

The higher inedibility with increasing prey diversity is supported by a recent meta-analysis revealing that consumer impacts on prey populations tend to decline with prey diversity (Hillebrand and Cardinale 2004).

In the only other study that directly manipulated bacterial diversity to examine decomposition responses, Wohl et al. (2004) found that increasing bacterial diversity increased cellulose decomposition. Because maximum cellulose decomposition in diverse bacterial communities exceeded that of single bacterial species, Wohl et al. (2004) concluded that complementarity was primarily responsible for the observed positive diversity effect. The importance of selection effects, however, cannot be readily determined as bacterial compositions were not replicated in their experiment, though they were likely to be of small magnitude owing to the use of bacterial species exhibiting similar cellulolytic activities (translating into similar competitive abilities with cellulose being the main carbon source). In the present study, bacterial species differed significantly in both competitive ability and decomposing capacity, and competitively dominant species did not contribute dominantly to decomposition; as a result little complementarity effects coupled with negative selection effects led to decomposition being largely independent of bacterial diversity. This suggests that negative selection effects may be potentially common when examining the relationship between bacterial diversity and decomposition in natural bacterial communities or synthetic bacterial communities consisting of randomly selected species, where species competitive ability varies but may not necessarily correlate with species decomposition capacity.

Consistent with the common stimulatory role of bacterial consumption in carbon mineralization (e.g., Fenchel and Harrison 1976, Sherr et al. 1982), elevated decomposition was found in the presence of *Tetrahymena*. Proposed mechanisms for enhanced decomposition under bacterial consumption include consumer recycling of limiting nutrients, consumer secretion of bacterial growth-promoting substances, and consumer alteration of bacterial community structure (Fenchel and Harrison 1976, Ratsak et al. 1996). My experiment did not allow for direct examination of these mechanisms, but the fact that *Tetrahymena* consumption generally reduced the density of the best bacterial decomposer (Fig. 2) and promoted decomposition in both monocultures and polycultures suggests that consumer-induced changes in bacterial community structure cannot explain the observed pattern. Intriguingly, decomposition demonstrated a strong positive relationship with consumer abundance (Fig. 3). This result, if general, would suggest that decomposition may proceed at a more rapid pace in natural communities containing more abundant bacterivorous consumers. Note that this pattern is consistent with increased bacterial turnover rate due either to increased nutrient cycling or growth substance secretion by consumer populations. Several studies have reported increased organic matter decomposition with increasing food web diversity, without identifying underlying mechanisms (McGrady-Steed et al. 1997, Krumins et al. 2006). A possible explanation, based on the finding of this study, is that increasing diversity might have led to increases in bacterivorous consumer abundance, resulting in elevated decomposition in more complex food webs.

The presence of bacterivorous consumers did not affect the bacterial BEF relationships, regardless of the ecosystem variable examined. This finding contrasts with several experiments reporting that trophic interactions can alter the form of the BEF relationship (Mulder et al. 1999, Naeem et al. 2000, Duffy et al. 2005, Gamfeldt et al. 2005). For example, Mulder et al. (1999) found that insect herbivory reduced the positive effects of terrestrial plant diversity on primary production, and Duffy et al. (2005) found that blue crab (*Callinectes sapidus*) predation promoted a positive relationship between mesograzer diversity and secondary production in seagrass ecosystems. In both studies, herbivory/predation-induced changes in species relative abundance contributed to changes in the BEF relationship. In the present study, however, the bacterivorous consumer did not significantly alter the relative abundance of component species in polycultures; in particular, it did not affect the dominance of the superior bacterial competitor (Fig. 2). Thus the same mechanisms (positive or negative selection effects) that dominated in predation-free controls also dominated in the presence of consumers. Overall, the various (positive, negative, or little) effects of trophic interactions on the BEF relationship, which are also predicted by theoretical models (Holt and Loreau 2002, Thebault and Loreau 2003), illustrate the difficulty associated with predicting ecosystem responses to changes in biodiversity in multitrophic systems.

Although the presence of consumers did not, in general, alter the form of BEF relationships, apparent competition among bacterial species did cause small deviations from the general trend. Here apparent competition caused some bacterial polycultures to attain smaller community biovolumes than corresponding monocultures (Fig. 1B), resulting in negative selection effects in these treatments, contrasting with positive selection effects found for other treatments (Table 1). Apparent competition has long been recognized as a potentially important mechanism affecting species abundance and community structure (Holt 1977). This study indicates that apparent competition affects the BEF linkage, although it only occurred in a few treatments here without major consequences for the general BEF relationship. Given a growing body of evidence for apparent competition in a variety of habitats (Chaneton and Bonsall 2000), future biodiversity studies should consider apparent competition as a potential mechanism contributing to the BEF relationship in multi-trophic systems.

To summarize, increasing bacterial diversity had a positive effect on bacterial community biovolume, but did not affect decomposition of organic matter and consumer abundance. Positive selection effects underlay the positive bacterial-diversity–community-biovolume relationship, whereas negative selection effects decoupled decomposition and consumer abundance from bacterial diversity. That the form of BEF relationship differed for different ecosystem variables, a finding also reported by several other studies (e.g., Naeem et al. 1994, McGrady-Steed et al. 1997), suggests that the focus of current biodiversity studies on aggregate community biomass may not yield insights into how other ecosystem processes respond to changes in biodiversity. This stresses the need for future studies to broaden the spectrum of ecosystem variables examined. That negative selection effects rendered decomposition and consumer abundance being independent of bacterial diversity is consistent with the idea that negative selection effects may frequently contribute to BEF relationships when examining ecosystem functions other than aggregate community biomass. This result is likely to be robust given that there is no a prior reason to believe that bacterial traits associated with decomposing organic matter or sustaining consumer populations will be the same as those associated with species competitive ability. The limited number of bacterial BEF experiments conducted so far means that we have much work to do to confirm (or refute) the generality of this result.

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